

## Short communication. Divergent mass selection for different flowering times in a Spanish synthetic maize population

J. I. Ruiz de Galarreta<sup>1</sup> and A. Álvarez<sup>2\*</sup>

<sup>1</sup> NEIKER-Instituto Vasco de Investigación y Desarrollo Agrario. P.O. Box 46. 01080 Vitoria. Spain

<sup>2</sup> Estación Experimental de Aula Dei (CSIC). P.O. Box 202. 50080 Zaragoza. Spain

### Abstract

Eight cycles of divergent mass selection for flowering time were performed in the synthetic maize (*Zea mays* L.) population Lazcano (EVS9) from Guipuzcoa, Spain. A trial with a randomised block design plus three replicates was then performed at two locations in northeastern Spain, and over two years, to compare flowering time in the original population and the material produced in the eight selection cycles. Linear regression coefficients were determined to estimate the average rate of selection response per cycle. Genetic gains with respect to the vegetative cycle and other major agronomic traits were obtained. Direct selection responses were significant and grain yield maintained. No negative correlated responses were obtained for any agronomic trait. Divergent mass selection for early flowering was therefore found to be efficient in this synthetic population.

**Additional key words:** earliness, genetic gain, *Zea mays* L.

### Resumen

#### Comunicación corta. Selección masal divergente para precocidad en una variedad sintética española de maíz

Se llevaron a cabo ocho ciclos de selección masal divergente para precocidad en la población sintética de maíz (*Zea mays* L.) Lazcano (EVS9), originaria de Guipúzcoa, España. Se realizó un ensayo en dos localidades del Noreste de España y durante dos años, con un diseño de bloques completos al azar y tres repeticiones, con el fin de comparar el periodo de floración en la población original y los ocho ciclos de selección. Se han determinado los coeficientes de correlación lineal para estimar la respuesta media por ciclo de selección. Asimismo, se han calculado las ganancias genéticas para el ciclo vegetativo y los principales caracteres agronómicos. La respuesta directa a la selección fue significativa, manteniéndose el rendimiento. No se encontraron respuestas negativas correlacionadas con otros caracteres agronómicos. La selección divergente realizada para precocidad fue, por lo tanto, eficiente en esta población sintética.

**Palabras clave adicionales:** ganancia genética, precocidad, *Zea mays* L.

The unpredictability of climatic conditions underscores the need to improve the productivity and stability of crops under a wide range of environmental situations (Landi *et al.*, 2001). The landraces of maize (*Zea mays* L.) have long been cultivated in different environments, and have been subject to natural and human selection and to different cultivation practices. These landraces show resistance to pests and diseases, good quality grain, and have become adapted to different environmental conditions. Since they provide an important source of genetic variation for use in breeding programs, it is essential to obtain reliable information on these adapted populations.

Selection for early flowering increases the genetic diversity of maize. Early maturing inbreds and hybrids can be obtained from elite, late-maturing germplasm (Troyer, 1990). The maturity ratings for the maize usually grown in Aragón (Zaragoza, Huesca and Teruel) range from FAO 400 to 700 (Elicegui, 1990). Later maturing genotypes show high grain moisture levels at harvest and present problems with storage (Ordás, 1988). Therefore, early flowering is a desirable characteristic in humid and cold areas. Earliness is seen in local varieties, the product of long adaptation periods (Moreno-González, 1981). Selection for early flowering in late synthetic populations has been attempted in temperate zones to improve their adaptation (Ordás, 1988).

Mass selection, the oldest method employed in maize breeding, is simple and fast, but its effectiveness depends

\* Corresponding author: [jiruiz@neiker.net](mailto:jiruiz@neiker.net)

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on the heritability of the selected trait (Gardner, 1961). Stratified mass selection has been useful for improving grain yield, prolificacy, earliness, ear height, ear length and resistance to insects (Hallauer and Miranda, 1981; Ordás *et al.*, 1996; Weyhrich *et al.*, 1998). Early flowering has also shown good responses to mass selection. Hallauer and Sears (1972), Troyer and Brown (1976), Rubino and Daws (1990) and many other authors have reported the efficacy of this type of selection and have described the gains obtained by selection cycle in different populations. The aim of the present study was to evaluate divergent mass selection for flowering time in the material of eight selection cycles of a Spanish synthetic maize population.

The synthetic population Lazkano was developed at the Aula Dei Experimental Station in Zaragoza, Spain. It was obtained by crossing 19 old, Spanish, open-pollinated landraces from Lazkano and Ataun in Guipuzcoa (northern Spain). The crosses required to form the new synthetics were maintained in 1989 for one generation by bulk sibbing and then chain-crossed and bulked to provide a base population. The synthetics were intermated twice, and later labelled as cycle 0 (C0). One cycle of intrapopulation recurrent selection based on  $S_1$  lines was conducted for the C0 population in 1991–1993 before mass selection for flowering time began in Zaragoza in 1994.

Divergent mass selection was performed by selecting the 100 earliest and latest flowering plants (in terms of first silks) from a total sample of 1,000 plants. The selected plants were recombined by bulking pollen from the selected plants only. The planting density was 66,000 plants  $\text{ha}^{-1}$ .

A balanced bulk of seed from the recombined plants was used for the next cycle of mass selection. Selection in subsequent cycles was conducted in a similar way. After eight cycles of divergent selection, seed from the original population (C0) and from the material representing early flowering (early cycles, C1E to C8E) and late flowering maize (late cycles, C1L to C8L) were sib-mated in 2002 to eliminate the effect of seed age before their comparison in performance trials.

Field trials were undertaken at Arkaute, in the province of Álava ( $2^\circ 72' \text{W}$ ;  $42^\circ 83' \text{N}$ , altitude 550 m average rainfall 800 mm) and at Montañana, in the province of Zaragoza ( $0^\circ 47' \text{W}$ ;  $41^\circ 44' \text{N}$ , altitude 220 m, average rainfall 300 mm), in 2003 and 2005. Experiments were performed following a randomised complete-block design with three replicates. All trial material was machine-planted in May at 66,000 plants  $\text{ha}^{-1}$  at both sites.

Each plot was composed of two 5 m rows spaced 0.75 m apart. Plants at both localities were manually harvested in October. The cycles were considered fixed effects and the location as random. Five hybrid checks were included in the trials: CM105  $\times$  CM109, A639  $\times$  F212, Stefania, Florencia and Natalia. Cultivation, fertilization and pest and weed control were carried out according to local practices.

Flowering (pollen shedding and days to silking) was checked daily and recorded as having occurred when 50% of the total plants in a plot had flowered. Days to flowering was calculated as the time elapsed between the emergence and flowering dates; average heat unit accumulation was determined from the number of days above  $10^\circ \text{C}$  in the same period. Plant height (cm), ear height (cm), total leaf number and ear node number were measured after flowering. The mean kernel moisture for the total plot was determined at harvest using an electronic meter ( $\text{g H}_2\text{O kg}^{-1}$ ). Lodging was estimated as the percentage of plants showing either root or stalk lodging (%). The grain yield for the entire plot was calculated as the weight of grain expressed as  $\text{Mg ha}^{-1}$ , adjusted for a kernel moisture of  $140 \text{ g H}_2\text{O kg}^{-1}$ . Other traits evaluated included ear length (mm), ear diameter (mm), the number of kernels per ear, and 1,000 kernel weight (g).

ANOVA and the homogeneity variance test (Bartlett, 1947) were performed for each assay before combining assays. Means were compared by the LSD method (Steel and Torrie, 1980). All calculations were performed using the SAS package (SAS, 2000). The linear regression coefficients obtained from the regression models were used as estimates of the average rate of response per cycle. Linear regressions were obtained using the PROC GLM and PROC CORR procedures respectively (SAS, 2000).

Individual analyses of variance showed highly significant differences for flowering traits between late and early material across locations (data not shown). In Montañana, significant differences were observed between them for grain moisture, lodging, yield and 1,000 seed weight. In Arkaute, highly significant differences were found for grain yield and 1,000 seed weight. No significant differences were detected for any other plant or agronomic traits.

Combined ANOVA showed highly significant differences between the late and early material with respect to days to flowering and average heat unit accumulation; the interaction *cycle*  $\times$  *locality* also had a significant effect (data not shown). The result of the Bartlett test

was significant; therefore, mean comparisons for each trait were made using individual values for each locality. No significant differences were found with respect to the factor «year» for any of the evaluated traits. Differences between late and early cycles were seen across locations in terms of grain moisture at harvest, 1,000 seed weight, and grain yield. Significant differences between cycles were detected at both locations for plant and ear traits (except for the number of ear rows).

Increases in the number of days to pollen shed and days to silking were generally consistent after selecting for later maturity (Table 1). However, reductions in

these times were obtained when selection for early maturity was undertaken. Grain yield increased after selection for later maturing genotypes.

Table 2 shows the linear regression coefficients for the traits evaluated in both locations and highlights the highly significant genetic gains obtained (pollen shed, silking, heat units to pollen shed and heat units to silking) in late and early flowering. The gain for days to silking was 0.37 and 0.90 d cycle<sup>-1</sup> for the late and early cycles respectively in the Arkaute trial, and 0.36 and 0.58 in Montañana. In both trials, the number of heat units accumulated differed significantly in both the late and early flowering cycle material. A significant

**Table 1.** Means trait values for 2003 and 2005 in late and early cycles of the synthetic Lazkano maize population in Arkaute and Montañana

| Traits                               | Arkaute |      |      |      |      |               | Montañana |      |      |      |      |               |
|--------------------------------------|---------|------|------|------|------|---------------|-----------|------|------|------|------|---------------|
|                                      | C0      | C2   | C4   | C6   | C8   | LSD<br>(0.05) | C0        | C2   | C4   | C6   | C8   | LSD<br>(0.05) |
| <i>Late cycles</i>                   |         |      |      |      |      |               |           |      |      |      |      |               |
| Pollen shed (days)                   | 84      | 85   | 87   | 88   | 89   | 2.30          | 67        | 68   | 68   | 68   | 70   | 1.29          |
| Silking (days)                       | 85      | 87   | 91   | 90   | 92   | 2.60          | 70        | 71   | 71   | 70   | 72   | 1.47          |
| Heat units to pollen shed (°C)       | 649     | 663  | 671  | 685  | 697  | 12.5          | 710       | 719  | 728  | 728  | 750  | 14.1          |
| Heat units to silking (°C)           | 665     | 681  | 710  | 708  | 722  | 13.4          | 750       | 764  | 764  | 749  | 772  | 15.0          |
| Grain yield (Mg ha <sup>-1</sup> )   | 4.71    | 5.60 | 6.90 | 7.52 | 8.19 | 0.73          | 3.01      | 5.98 | 8.89 | 9.02 | 8.91 | 0.67          |
| Lodging (%)                          | 21.2    | 17.2 | 16.9 | 30.1 | 20.4 | 7.21          | 27.2      | 15.1 | 18.9 | 6.5  | 8.7  | 4.97          |
| Grain moisture (g kg <sup>-1</sup> ) | 334     | 309  | 305  | 296  | 285  | 39.1          | 198       | 210  | 205  | 211  | 237  | 13.1          |
| 1,000-weight seed (g)                | 434     | 426  | 440  | 465  | 443  | 16.3          | 352       | 331  | 350  | 331  | 342  | 14.7          |
| Ear length (cm)                      | 16.0    | 17.1 | 15.8 | 16.8 | 18.4 | 1.30          | 16.8      | 16.9 | 17.0 | 18.3 | 17.5 | 1.30          |
| Ear diameter (mm)                    | 48.9    | 49.0 | 48.9 | 49.5 | 49.6 | 3.11          | 43.5      | 45.0 | 46.8 | 47.3 | 44.9 | 2.48          |
| Kernel row number                    | 12.7    | 13.1 | 13.1 | 13.1 | 13.4 | 0.46          | 12.0      | 12.5 | 13.3 | 13.4 | 14.0 | 0.53          |
| Plant height (cm)                    | 230     | 253  | 238  | 249  | 245  | 20.0          | 149       | 153  | 154  | 162  | 187  | 16.3          |
| Ear height (cm)                      | 93      | 112  | 95   | 101  | 104  | 11.3          | 67        | 69   | 71   | 71   | 80   | 9.60          |
| Number of leaves                     | 13.0    | 14.0 | 12.0 | 13.1 | 13.1 | 1.42          | 10.0      | 11.0 | 11.1 | 11.0 | 12.0 | 1.21          |
| Ear node                             | 8.0     | 9.2  | 8.0  | 8.2  | 8.2  | 1.20          | 6.0       | 6.1  | 6.5  | 6.2  | 6.7  | 1.06          |
| <i>Early cycles</i>                  |         |      |      |      |      |               |           |      |      |      |      |               |
| Pollen shed (days)                   | 83      | 84   | 81   | 81   | 80   | 2.27          | 67        | 66   | 65   | 65   | 64   | 1.20          |
| Silking (days)                       | 85      | 86   | 85   | 83   | 83   | 2.42          | 70        | 69   | 69   | 68   | 67   | 1.37          |
| Heat units to pollen shed (°C)       | 641     | 652  | 632  | 628  | 614  | 9.8           | 710       | 700  | 681  | 683  | 675  | 10.3          |
| Heat units to silking (°C)           | 665     | 673  | 664  | 650  | 648  | 12.1          | 750       | 741  | 739  | 719  | 705  | 13.1          |
| Grain yield (Mg ha <sup>-1</sup> )   | 4.70    | 5.01 | 7.04 | 7.54 | 6.90 | 0.78          | 3.98      | 5.01 | 7.12 | 7.92 | 7.80 | 0.45          |
| Lodging (%)                          | 20.4    | 25.3 | 27.1 | 16.7 | 16.0 | 5.84          | 27.3      | 25.3 | 26.7 | 18.5 | 22.2 | 8.34          |
| Grain moisture (g kg <sup>-1</sup> ) | 340     | 329  | 265  | 328  | 310  | 50.5          | 201       | 226  | 230  | 210  | 207  | 16.2          |
| 1,000-weight seed (g)                | 432     | 400  | 445  | 455  | 417  | 15.0          | 354       | 343  | 345  | 353  | 357  | 7.2           |
| Ear length (cm)                      | 15.8    | 16.8 | 17.1 | 16.9 | 16.1 | 1.20          | 17.0      | 17.9 | 18.0 | 17.1 | 17.0 | 1.21          |
| Ear diameter (mm)                    | 48.6    | 48.1 | 47.9 | 48.0 | 47.5 | 3.20          | 43.7      | 44.2 | 41.9 | 43.3 | 43.0 | 2.18          |
| Kernel row number                    | 12.5    | 13.0 | 12.4 | 12.8 | 13.0 | 0.43          | 12.0      | 13.0 | 12.3 | 12.0 | 11.7 | 0.44          |
| Plant height (cm)                    | 230     | 243  | 246  | 235  | 232  | 22.3          | 149       | 147  | 160  | 155  | 149  | 10.7          |
| Ear height (cm)                      | 93      | 100  | 105  | 88   | 92   | 11.6          | 65        | 70   | 81   | 72   | 68   | 13.7          |
| Number of leaves                     | 13.2    | 13.4 | 14.0 | 12.6 | 12.3 | 1.22          | 10.3      | 10.8 | 11.2 | 11.1 | 10.3 | 0.22          |
| Ear node                             | 8.3     | 8.5  | 9.1  | 8.3  | 8.1  | 1.40          | 5.6       | 6.1  | 7.4  | 6.6  | 5.3  | 0.42          |

**Table 2.** Linear regression coefficients for traits of maize selected for late and early flowering in the Arkaute and Montañana trials

| Traits                               | Arkaute                  |                           | Montañana                |                           |
|--------------------------------------|--------------------------|---------------------------|--------------------------|---------------------------|
|                                      | Late cycles <sup>1</sup> | Early cycles <sup>2</sup> | Late cycles <sup>1</sup> | Early cycles <sup>2</sup> |
| Pollen shed (days)                   | 0.78**                   | -0.49**                   | 0.38**                   | -0.34**                   |
| Silking (days)                       | 0.90**                   | -0.37**                   | 0.58**                   | -0.36**                   |
| Heat units to pollen shed (°C)       | 6.70**                   | -4.42**                   | 5.55**                   | -4.59**                   |
| Heat units to silking (°C)           | 7.35**                   | -3.27**                   | 1.41**                   | -5.68**                   |
| Grain yield (Mg ha <sup>-1</sup> )   | 0.40**                   | 0.20                      | 0.71**                   | 0.58                      |
| Lodging (%)                          | 0.83                     | -1.96                     | -2.07*                   | -0.81                     |
| Grain moisture (g kg <sup>-1</sup> ) | -0.23                    | -0.29                     | 0.06                     | 0.08                      |
| 1,000-weight seed (g)                | 3.29                     | 0.91                      | -1.00                    | -0.61                     |
| Ear length (cm)                      | 1.57                     | -0.90                     | 2.04*                    | -1.15                     |
| Ear diameter (mm)                    | 0.02                     | -0.26                     | 0.25                     | -0.10                     |
| Kernel row number                    | 0.07                     | -0.03                     | 0.17                     | -0.08                     |
| Plant eight (cm)                     | 1.35                     | -1.00                     | 0.23                     | 0.28                      |
| Ear height (cm)                      | 0.12                     | -1.84                     | 0.58                     | 0.01                      |
| Number of leaves                     | 0.01                     | -0.11                     | 0.21*                    | 0.01                      |
| Ear node                             | -0.05                    | -0.06                     | 0.08                     | 0.01                      |

<sup>1</sup> Maize selected for late flowering. <sup>2</sup> Maize selected for early flowering. \* P < 0.05. \*\* P < 0.01.

reduction of 3.27 heat units cycle<sup>-1</sup> was recorded for the early cycle material, and an increase of 7.35 units for the late cycle material in the Arkaute trial. In the Montañana trial, a reduction of 5.6 heat units cycle<sup>-1</sup> was recorded for the early cycle material, and an increase of 1.41 heat units cycle<sup>-1</sup> was seen for the late cycle material. For grain yield, a linear trend with a gain of 0.40 Mg ha<sup>-1</sup> was observed in Arkaute, while a genetic gain of 0.71 Mg ha<sup>-1</sup> was observed in Montañana.

Bletsos and Goulas (1999) found an average response of 5.1% per cycle for grain yield in a single cross hybrid after three cycles of mass selection for increasing grain protein concentration. Ordás (1988) reported reductions in grain yield in American populations selected for early flowering. Subandi (1985) and Troyer and Larkins (1985) reported similar results. However Fonturbel and Ordás (1981) found higher gains when selecting for earliness in the populations Purdue A and Purdue B.

The present preliminary results confirm that mass selection continues to be an efficient method for highly heritable traits. A positive response to selection was obtained for both early and late flowering after eight cycles of selection. Selection for late flowering was associated with an expected genetic gain for grain yield. The improved plants developed in this divergent mass selection program could be released as sources of inbred lines to produce hybrids adapted to different environmental conditions.

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